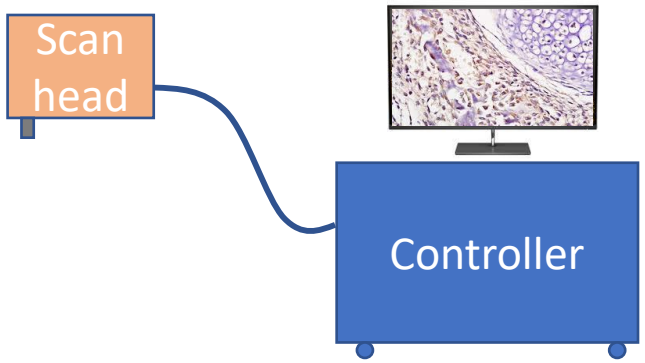
Next Generation Multi-Modal Imaging

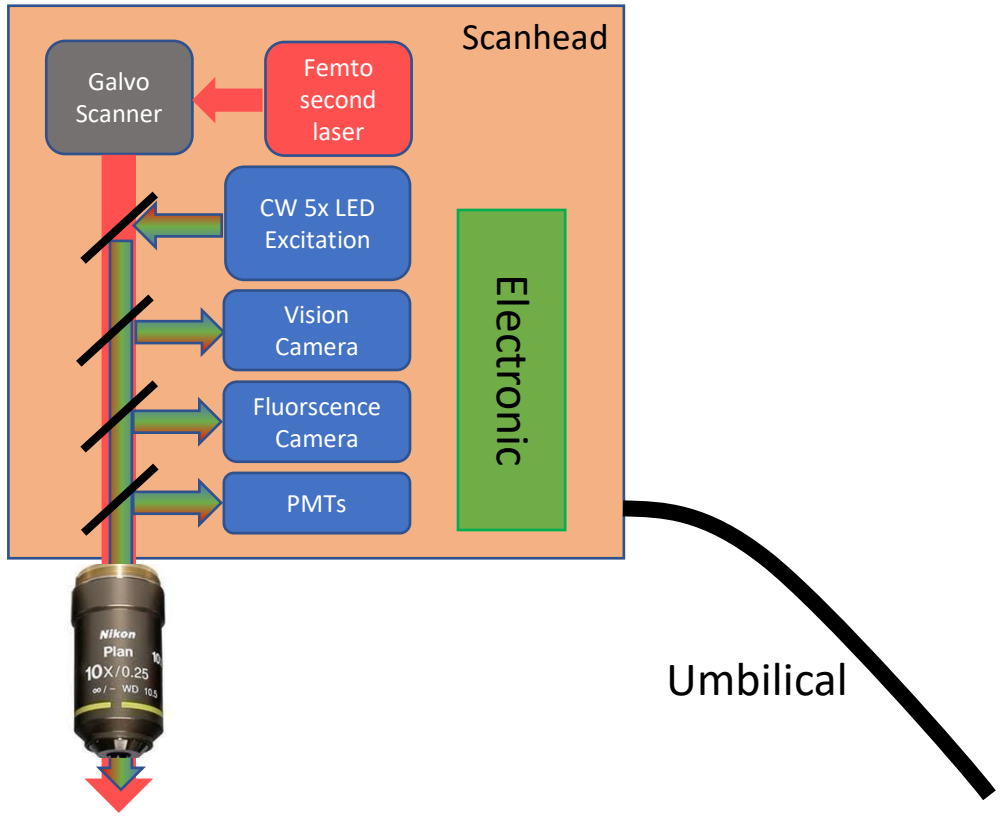
EPIC Online Technology Meeting on in-vivo Imaging

Dr. Christof Böhler, May 04-2020

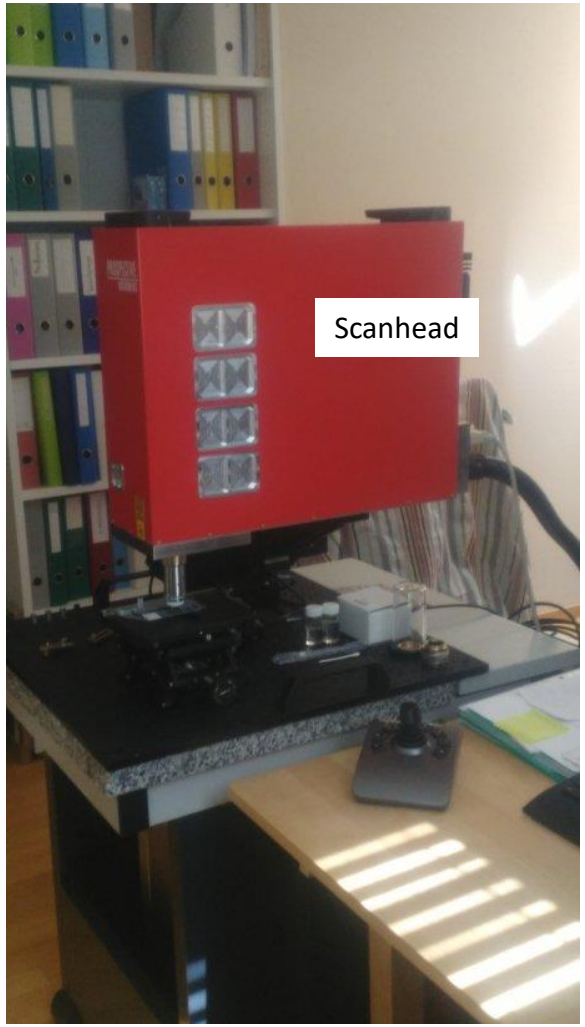
Current multimodal imaging platform MP2-1030

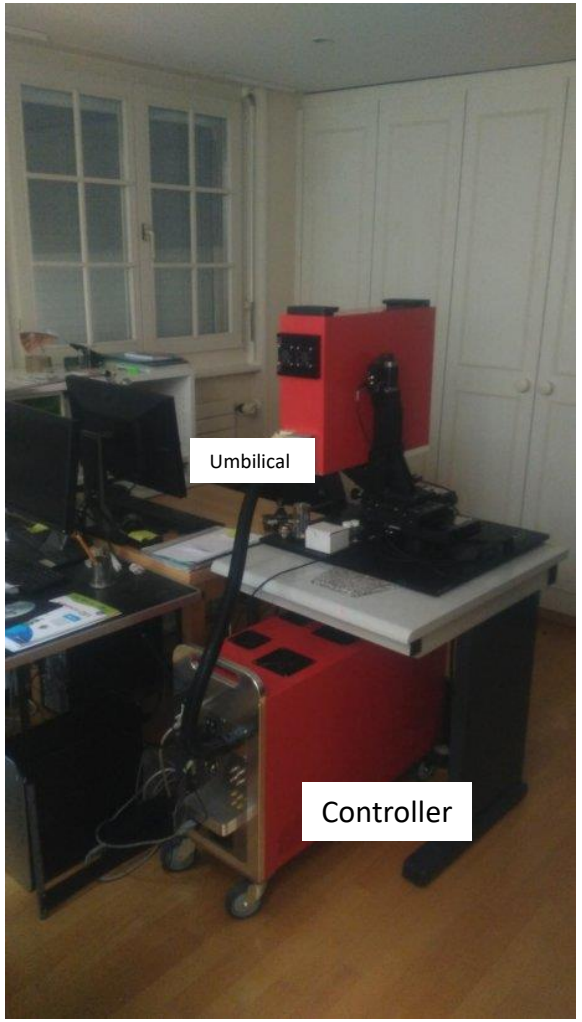
1. Combining complementary modalities into one compact and easy to use device:
   Multiphoton (SHG/THG/2PE; fs laser + PMTs), Fluorescence imaging (camera + LED excitation), Vision camera (brightfield).
2. Freely-moveable scan-head, all-integrated, no alignment, easy to operate and to install (even PC included).
3. Label-free, no tissue preparation, little photo-toxicity.
4. Mosaic imaging: from mm to cm scale (stitching).
5. Epi-detection in backscatter direction

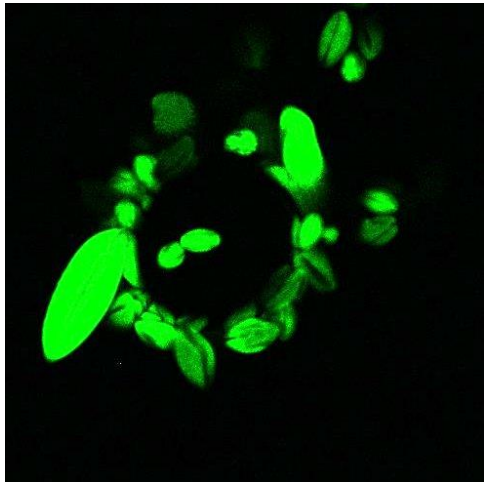




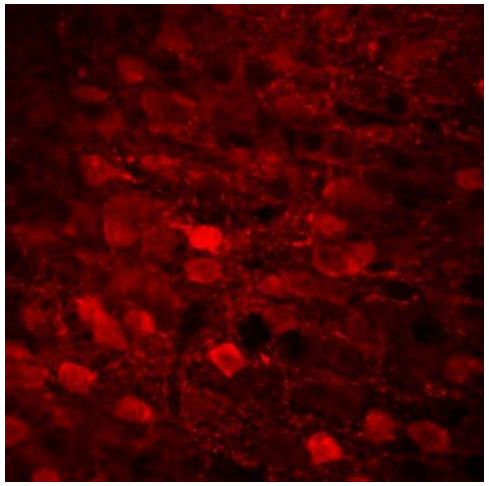
Works after transport in the home office ☺





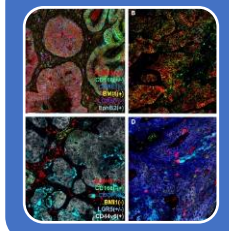


Pollen, SHG



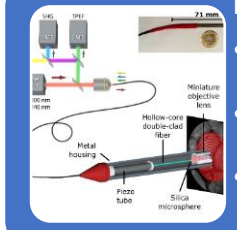
Neurons, 2PEF

What applications in imaging?



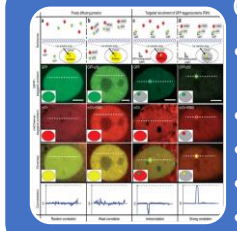
Routine surgery

- Fast optical biopsies on a microscope in the OT replacing H/E staining procedure => instant pathology
- Define tumor boundaries
- Automatic image analysis
- Tumor micro environment
- What does the surgeon need to see? Which modalities?



Endoscopy imaging

- What about a fibre based microscope built in the endoscope probe?
- https://www.reboundmd.com/news/what-endoscopic-brain-surgery
- Prostate cancer tumor boundaries

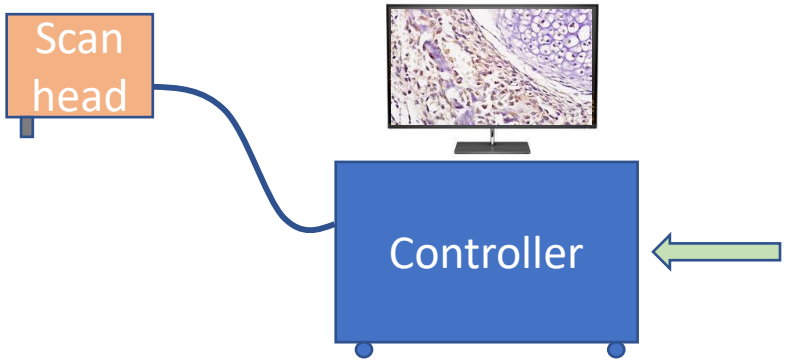


Clinical Research => outlook

- Molecular and ultra-structural Imaging => drug-target interactions => responders/non-responders
- SHG/THG/2PE plus Raman (CARS, SRS,)
- Opto-pharmacology
- AI, image processing, deep learning
- Opto-Genetics

APPLICATIONs currently envisaged

- Oncology research => future in clinic use
- Neurology research, => future in clinic use
- Dermatology clinical research

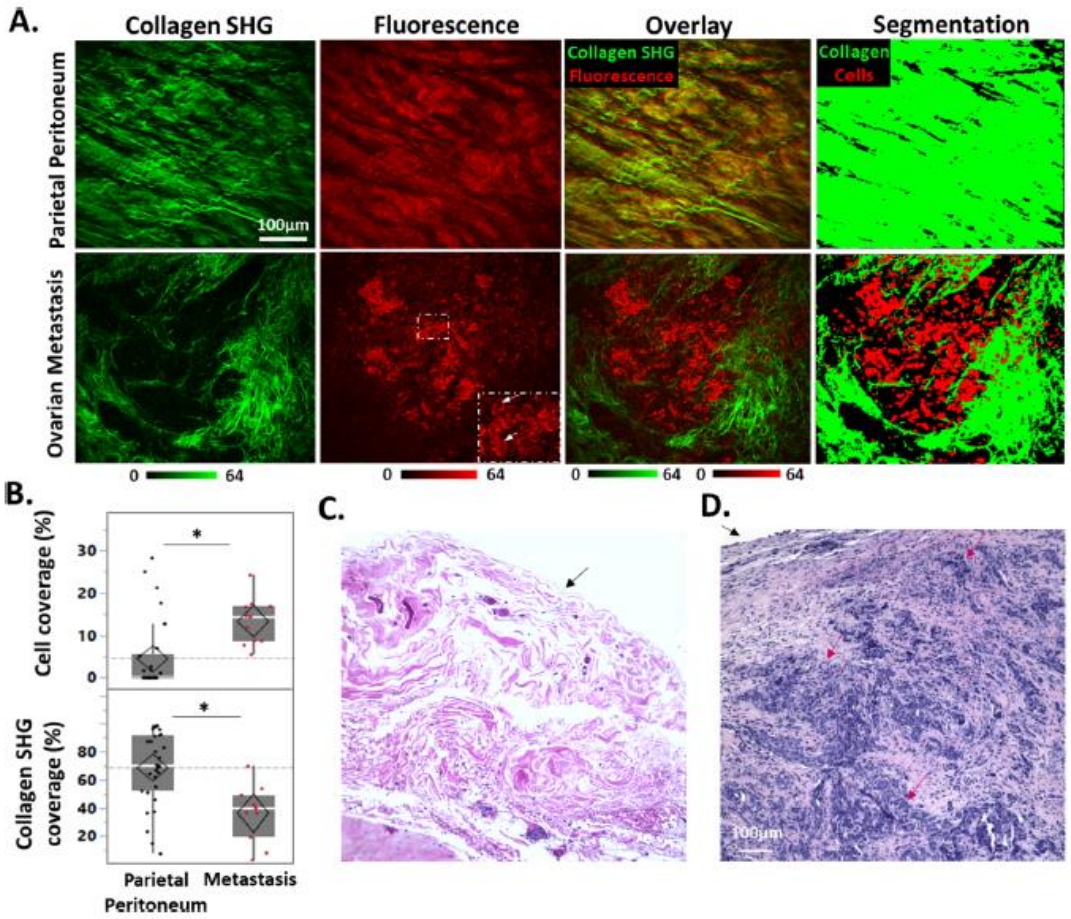


GOALs

- Decide which modalities are «must haves»
- Provide «can have» modalities on demand

Example Oncology – metastasis visualisation

www.ncbi.nlm.nih.gov/pmc/articles/PMC6757455/ , Thomas Schnelldorfer, thomas.schnelldorfer@lahey.org, Bedford MA



Biopsy taken → Put on microscope → Interpret images

Instant pathology – label-free- results in minutes

Fig. 1.

Cancer invasion alters tissue component constitution **A.** Examples of two-photon label-free images capturing SHG (pseudocolored green) and autofluorescence (pseudocolored red) signals for healthy parietal peritoneum and ovarian metastatic tissue. Signal overlay reveals spatial overlap of collagen SHG and fluorescence in healthy tissues, whereas destruction of matrix components by cancerous cellular infiltration is observed in metastasis. Insert in ovarian metastasis fluorescence image highlights the presence of cellular clusters (white arrows), identified by dark nuclei and bright cytoplasmic features. **B.** Quantification of image area coverage by automated image feature segmentation as shown in A. *denotes significance at a = 0.05 **C-D**. Examples of histological H&E microscopic images for each tissue group (C-Healthy,D-Metastasis). Black arrows indicate mesothelial surface while red arrows highlight infiltrative cancerous cellular clusters in the metastatic biopsy. Mesothelial cells were not typically recognized in the examined sections.

Example Dermatology – SGH, 2PEF, CARS

https://pubs.rsc.org/en/content/articlelanding/2019/pp/c8pp00410b#!divAbstract , angelika.unterhuber@meduniwien.ac.at

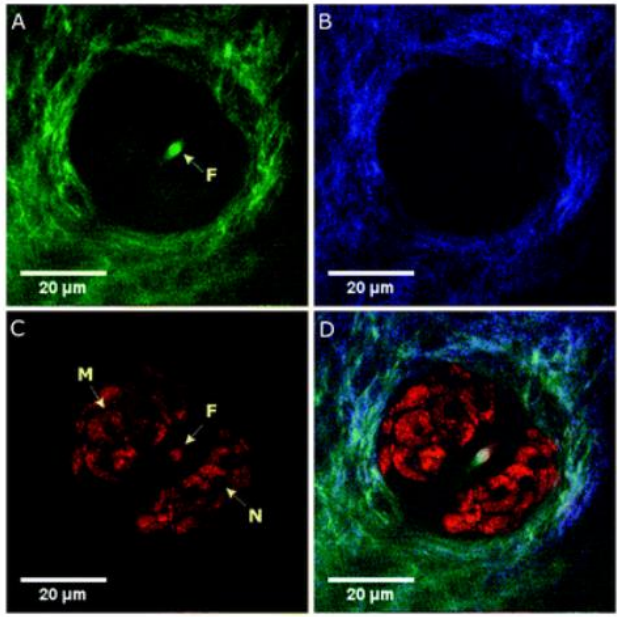


Fig. 3 Epi-detected multimodal NLOI images of a sebaceous gland. (A) Elastin and (B) collagen content of the ECM supporting the gland visualized through TPEF and SHG, respectively; (C) lipid content of the gland imaged by CARS where it is possible to identify the sebocytes' membranes (M) and nuclei (N) and as well the location of the follicle (F). (D) Merged image of (A), (B) and (C) showing the complementary information provided by TPEF, SHG and CARS.

Paper: Depth resolved label-free multimodal optical imaging platform to study morpho-molecular composition of **dermal tissue**

Multimodal imaging platforms offer a vast array of tissue information in a single image acquisition by combining complementary imaging techniques. By merging different systems, better tissue characterization can be achieved than is possible by the constituent imaging modalities alone. The combination of optical coherence tomography (OCT) with non-linear optical imaging (NLOI) techniques such as two-photon excited fluorescence (TPEF), second harmonic generation (SHG) and coherent anti-Stokes Raman scattering (CARS) provides access to detailed information of tissue structure and molecular composition in a fast, label-free and non-invasive manner. We introduce a multimodal label-free approach for morpho-molecular imaging and spectroscopy and validate the system in mouse skin demonstrating the potential of the system for co-localized acquisition of OCT and NLOI signals.

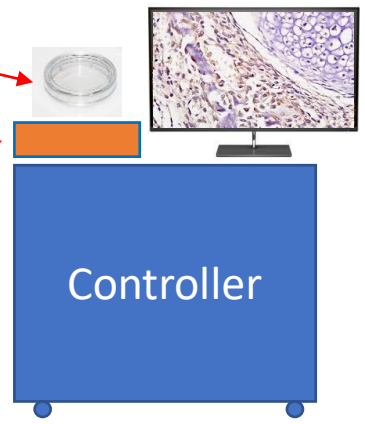
Multimodal medical-grade imaging platform

1. Automatic digital image staining and analysis by deep learning algorithm
2. Comparison of digital imaging with histopathology
3. Easy detection of tumor regression/disease progression
4. Combining imaging with spectroscopy, early detection of patient responsivness to therapy
5. Visualizing dynamic response of therapeutic modality on target
6. Fast, label-free, no tissue preparation, neglectable phototoxicity
7. Mosaic imaging: from µm to cm scale, large stitching
8. Ex-vivo small footprint device
9. In-vivo small, flexible tool: Bioptic needle
10. Epi-detection in backscatter direction

Ex-vivo:

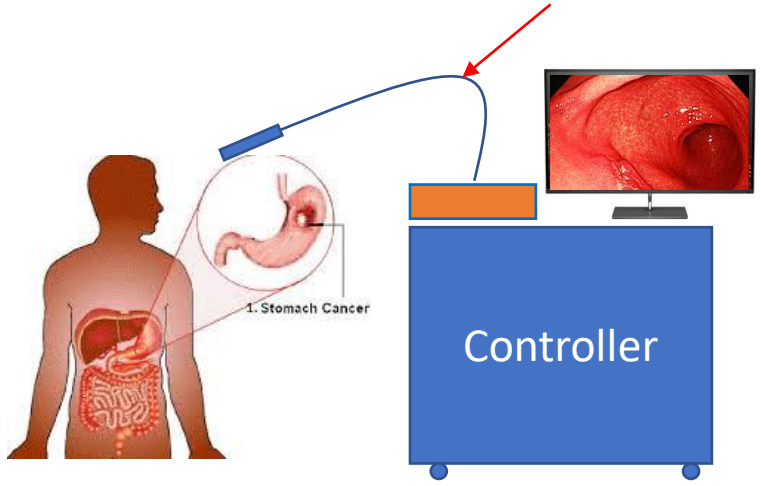
Fluorodish resected tissue holder

Inverted sample scanner



In-vivo:

Multimodal bioptic needle



PROSPECTIVE INSTRUMENTS